

SALINITY TOLERANCE AND BRANCHIAL CHLORIDE CELLS IN EURYHALINE TILAPIA (*Oreochromis mossambicus*)

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In teleost fish, plasma osmolality is maintained around 300 mOsm independent of environmental salinity. Branchial chloride cells (CCs) are involved importantly in the osmoregulatory processes, creating ionic and osmotic gradients between the body fluid and external environments. In particular, CCs are considered to be the salt-secreting site in seawater (SW) fish. The euryhaline tilapia, *Oreochromis mossambicus*, is able to adapt to a wide range of salinities from fresh water (FW) to concentrated SW. To clarify the involvement of CCs in the strong euryhalinity of the tilapia, we examined the morphological alteration of CCs in fish adapted to FW, SW and 180% SW. Although plasma osmolality increases slightly with increasing environmental salinity, the levels were maintained within a physiological range. Gill Na^+ , K^+ -ATPase activity became higher with increasing environmental salinity. CCs were more developed in SW than in FW, and most developed in 180% SW-adapted fish. Therefore, activated CCs in hyperosmotic environment are considered to be the site for salt secretion. The strong euryhalinity of the tilapia may be attributed to their excellent ability to develop CCs in response to increased environmental salinity.

HIGH SPEED VIDEO ANALYSIS OF BEAT PERIOD FLUCTUATION IN SEA-URCHIN EMBRYO CILIA

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It has been demonstrated that dopamine induces an increase in the fluctuation of beat period of sea-urchin embryo cilia and serotonin a decrease in it (Wada, Y., Mogami, Y. and Baba, S.A. (1997). J. Exp. Biol. 200:9-18). We found here that the membrane potential depolarization induced by elevated $[\text{K}^+]_o$ increased the fluctuation and hyperpolarization decreased it. The frame-by-frame analysis of ciliary beating pattern recorded with a high speed video camera showed that variation of the fluctuation under the influence of dopamine, serotonin and the membrane potential was mostly due to that in the length of pauses at the early and later phases of the effective stroke.

SYNCHRONOUS FIRING PATTERNS OF A SET OF INSECT NEURO-SECRETORY CELLS.

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The silk moth, *Bombyx mori*, has five pairs of neurosecretory cells releasing pheromoneotropic neuropeptides in the subesophageal ganglion. The neurosecretory cells send an axon to the corpus cardiacum via a branch (NCC-V) of the maxillary nerve. The cells usually fire within 20 ms; sometime a few cells and sometimes all the cells participate in a synchronous firing. Compound action potentials were recorded from left and right NCC-V and determine how many cells participating in each firing event. The probability curve as a function of number of firing cells was usually maximal at 1 cell (solitary firing), declined to a minimal value at 3-4 cells, and gradually increased with an increase in number of cells. There was no characteristic temporal feature in the time series of number of firing cells. A synchronous firing of multiple units is likely mediated by an electrical coupling among them. To reveal a coupling mechanism which initiates and accomplishes a synchronous firing of multiple units, the order of firing of a particular unit among five units was examined. Majority of firing events in the unit occurred in a particular order. Coupling mechanisms producing those characteristic firing patterns were discussed.

 Ca^{2+} CHANNEL ACTIVITY MODULATED BY COOLING OF THE LOBSTER CARDIAC MUSCLE

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Crustacean cardiac muscles often generate graded action potentials (spikes) on the excitatory junction potentials (EJPs). The spike amplitude increased in proportion to falling temperature in the cardiac muscle fibers of *Panulirus japonicus*. We have supposed that the spike potentials conform to inward currents of Ca^{2+} and that temperature acts on the Ca^{2+} channels. To examine these hypotheses, the muscle membrane currents were analyzed using voltage clamp methods and pharmacological agents in the nerve-muscle preparations of the lobster heart.

The muscle inward currents were divided into the rapid currents conforming to spike potentials; the slow currents to EJPs and the sustained currents induced by cooling. The former easily disappeared and the latter two hardly did with an application of Co^{2+} (10-20 mM). The rapid inward currents were blocked by nifedipine and also pertussis toxin (PTX). These data suggest that the cardiac muscle has L-type Ca^{2+} channels and that PTX-sensitive GTP-binding proteins (G proteins) concern the cold-sensitive Ca^{2+} currents. Then the G protein subunits, GDP- β -S and GTP- γ -S, were injected into the muscle fibers while the membrane currents were recorded. The GTP- γ -S enhanced the rapid currents while the GDP- β -S reduced them. This confirms that the G proteins regulate the Ca^{2+} channel function. Therefore, the enhancement of rapid Ca^{2+} currents may be resulting from cold suppression of G protein-mediated closing of the Ca^{2+} channels.

SENSORY BRISTLES ALONG THE WING MARGIN OF THE BUTTERFLY, *PIERIS RAPAE*.A. Yoshida¹, A. Noda¹ and J. Emoto². ¹JT Biohistory Res. Hall, Takatsuki, ²Biol. Lab., Nanzan Univ., Nagoya.

It was reported that sensory bristles are differentiated along the pupal wing margins in several moths (Clever, 1958). We morphologically and histochemically studied the sensory bristles along the adult wing margin of the butterfly, *Pieris rapae*. Results are as follows. 1) Sensory bristles are distributed along the larger part of the wing margin of *Pieris* than the moths previously reported. 2) The number of the bristles is about 60 in the forewing, and about 90 in the hindwing. All the bristles are completely covered by the pile of the hydrofuge scales. 3) The bristle is adjacent to fairly long marginal scales. 4) The external surface of the bristle lacks perforations. 5) Immediately near the bristle base, three large cellular nuclei are observed; it seems to be likely that they are the nuclei of the trichogen cell, the tormogen cell, and the sensory cell, respectively. Based on these results, we propose an airstream reception mechanism that these sensory bristles are stimulated by the marginal scale movement which is induced by an airstream.

CLASSIFICATION OF HEMOCYTES OF THE ASCIDIAN *HALOCYNTHIA RORETZI* BY FACS

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Hemocytes of the ascidian *Halocynthia roretzi* are believed to play an important role in self-defense system and allo-recognition. Since hemocytes consist of morphologically heterogeneous populations, it is essential to classify them and clarify the roles of each population in self-defense and allo-recognition. The hemocytes were classified into 12 groups by FACS using monoclonal antibodies and dyes. Our classification was compared to the morphological classifications previously reported.

Although tunic cells are thought to be a population of hemocytes that have migrated into the tunic, a portion of tunic cells show a unique feature, which has never found in hemocytes, by FACS analysis. The contact reaction was quantified by the increase in cell number of smaller population (cell debris) detected by FACS.